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CANCEROUS DISEASE MODIFYING ANTIBODIES

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REFERENCE TO RELATED APPLICATIONS

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FIELD OF THE INVENTION

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BACKGROUND OF THE INVENTION

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Each individual who presents with cancer is unique and has a cancer that is as different from other cancers as that person's identity. Despite this, current therapy treats all patients with the same type of cancer, at the same stage, in the same way. At least 30 percent of these patients will fail the first line of therapy, thus leading to further rounds of treatment and the increased probability of treatment failure, metastases, and ultimately, death. A superior

1 approach to treatment would be the customization of therapy for the particular individual. The
2 only current therapy that lends itself to customization is surgery. Chemotherapy and radiation
3 treatment cannot be tailored to the patient, and surgery by itself, in most cases is inadequate for
4 producing cures.

5 With the advent of monoclonal antibodies, the possibility of developing methods for
6 customized therapy became more realistic since each antibody can be directed to a single
7 epitope. Furthermore, it is possible to produce a combination of antibodies that are directed to
8 the constellation of epitopes that uniquely define a particular individual's tumor.

9 Having recognized that a significant difference between cancerous and normal cells is
10 that cancerous cells contain antigens that are specific to transformed cells, the scientific
11 community has long held that monoclonal antibodies can be designed to specifically target
12 transformed cells by binding specifically to these cancer antigens; thus giving rise to the belief
13 that monoclonal antibodies can serve as "Magic Bullets" to eliminate cancer cells.

14 Monoclonal antibodies isolated in accordance with the teachings of the instantly
15 disclosed invention have been shown to modify the cancerous disease process in a manner
16 which is beneficial to the patient, for example by reducing the tumor burden, and will
17 variously be referred to herein as cancerous disease modifying antibodies (CDMAB) or "anti-
18 cancer" antibodies.

19 At the present time, the cancer patient usually has few options of treatment. The
20 regimented approach to cancer therapy has produced improvements in global survival and

1 morbidity rates. However, to the particular individual, these improved statistics do not
2 necessarily correlate with an improvement in their personal situation.

3 Thus, if a methodology was put forth which enabled the practitioner to treat each tumor
4 independently of other patients in the same cohort, this would permit the unique approach of
5 tailoring therapy to just that one person. Such a course of therapy would, ideally, increase the
6 rate of cures, and produce better outcomes, thereby satisfying a long-felt need.

7 Historically, the use of polyclonal antibodies has been used with limited success in the
8 treatment of human cancers. Lymphomas and leukemias have been treated with human
9 plasma, but there were few prolonged remissions or responses. Furthermore, there was a lack
10 of reproducibility and no additional benefit compared to chemotherapy. Solid tumors such as
11 breast cancers, melanomas and renal cell carcinomas have also been treated with human blood,
12 chimpanzee serum, human plasma and horse serum with correspondingly unpredictable and
13 ineffective results.

14 There have been many clinical trials of monoclonal antibodies for solid tumors. In the
15 1980s there were at least 4 clinical trials for human breast cancer which produced only 1
16 responder from at least 47 patients using antibodies against specific antigens or based on tissue
17 selectivity. It was not until 1998 that there was a successful clinical trial using a humanized
18 anti-Her2 antibody in combination with cisplatin. In this trial 37 patients were accessed for
19 responses of which about a quarter had a partial response rate and another half had minor or
20 stable disease progression.

21 The clinical trials investigating colorectal cancer involve antibodies against both

1 glycoprotein and glycolipid targets. Antibodies such as 17-1A, which has some specificity for
2 adenocarcinomas, had undergone Phase 2 clinical trials in over 60 patients with only 1 patient
3 having a partial response. In other trials, use of 17-1A produced only 1 complete response and
4 2 minor responses among 52 patients in protocols using additional cyclophosphamide. Other
5 trials involving 17-1A yielded results that were similar. The use of a humanized murine
6 monoclonal antibody initially approved for imaging also did not produce tumor regression. To
7 date there has not been an antibody that has been effective for colorectal cancer. Likewise
8 there have been equally poor results for lung, brain, ovarian, pancreatic, prostate, and stomach
9 cancers. There has been some limited success in the use of an anti-GD3 monoclonal antibody
10 for melanoma. Thus, it can be seen that despite successful small animal studies that are a
11 prerequisite for human clinical trials, the antibodies that have been tested thus far, have been
12 for the most part, ineffective.

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14 PRIOR PATENTS

15 U.S. Patent No. 5,750,102 discloses a process wherein cells from a patient's tumor are
16 transfected with MHC genes which may be cloned from cells or tissue from the patient. These
17 transfected cells are then used to vaccinate the patient.

18 U.S. Patent No. 4,861,581 discloses a process comprising the steps of obtaining
19 monoclonal antibodies that are specific to an internal cellular component of neoplastic and
20 normal cells of the mammal but not to external components, labeling the monoclonal antibody,
21 contacting the labeled antibody with tissue of a mammal that has received therapy to kill

1 neoplastic cells, and determining the effectiveness of therapy by measuring the binding of the
2 labeled antibody to the internal cellular component of the degenerating neoplastic cells. In
3 preparing antibodies directed to human intracellular antigens, the patentee recognizes that
4 malignant cells represent a convenient source of such antigens.

5 U.S. Patent No. 5,171,665 provides a novel antibody and method for its production.
6 Specifically, the patent teaches formation of a monoclonal antibody which has the property of
7 binding strongly to a protein antigen associated with human tumors, e.g. those of the colon and
8 lung, while binding to normal cells to a much lesser degree.

9 U.S. Patent No. 5,484,596 provides a method of cancer therapy comprising surgically
10 removing tumor tissue from a human cancer patient, treating the tumor tissue to obtain tumor
11 cells, irradiating the tumor cells to be viable but non-tumorigenic, and using these cells to
12 prepare a vaccine for the patient capable of inhibiting recurrence of the primary tumor while
13 simultaneously inhibiting metastases. The patent teaches the development of monoclonal
14 antibodies which are reactive with surface antigens of tumor cells. As set forth at col. 4, lines
15 45 et seq., the patentees utilize autochthonous tumor cells in the development of monoclonal
16 antibodies expressing active specific immunotherapy in human neoplasia.

17 U.S. Patent No. 5,693,763 teaches a glycoprotein antigen characteristic of human
18 carcinomas is not dependent upon the epithelial tissue of origin.

19 U.S. Patent No. 5,783,186 is drawn to anti-Her2 antibodies which induce apoptosis in
20 Her2 expressing cells, hybridoma cell lines producing the antibodies, methods of treating
21 cancer using the antibodies and pharmaceutical compositions including said antibodies.

1 U.S. Patent No. 5,849,876 describes new hybridoma cell lines for the production of
2 monoclonal antibodies to mucin antigens purified from tumor and non-tumor tissue sources.

3 U.S. Patent No. 5,869,268 is drawn to a method for generating a human lymphocyte
4 producing an antibody specific to a desired antigen, a method for producing a monoclonal
5 antibody, as well as monoclonal antibodies produced by the method. The patent is particularly
6 drawn to the production of an anti-HD human monoclonal antibody useful for the diagnosis
7 and treatment of cancers.

8 U.S. Patent No. 5,869,045 relates to antibodies, antibody fragments, antibody
9 conjugates and single chain immunotoxins reactive with human carcinoma cells. The
10 mechanism by which these antibodies function is two-fold, in that the molecules are reactive
11 with cell membrane antigens present on the surface of human carcinomas, and further in that
12 the antibodies have the ability to internalize within the carcinoma cells, subsequent to binding,
13 making them especially useful for forming antibody-drug and antibody-toxin conjugates. In
14 their unmodified form the antibodies also manifest cytotoxic properties at specific
15 concentrations.

16 U.S. Patent No. 5,780,033 discloses the use of autoantibodies for tumor therapy and
17 prophylaxis. However, this antibody is an anti-nuclear autoantibody from an aged mammal. In
18 this case, the autoantibody is said to be one type of natural antibody found in the immune
19 system. Because the autoantibody comes from "an aged mammal", there is no requirement that
20 the autoantibody actually comes from the patient being treated. In addition the patent discloses
21 natural and monoclonal anti-nuclear autoantibody from an aged mammal, and a hybridoma cell

1 line producing a monoclonal anti-nuclear autoantibody.

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3 SUMMARY OF THE INVENTION

4 The instant inventors have previously been awarded U.S. Patent 6,180,357,
5 entitled "Individualized Patient Specific Anti-Cancer Antibodies" directed to a process for
6 selecting individually customized anti-cancer antibodies which are useful in treating a
7 cancerous disease. For the purpose of this document, the terms "antibody" and "monoclonal
8 antibody" (mAb) may be used interchangeably and refer to intact immunoglobulins produced
9 by hybridomas (e.g. murine or human), immunoconjugates and, as appropriate,
10 immunoglobulin fragments and recombinant proteins derived from immunoglobulins, such as
11 chimeric and humanized immunoglobulins, F(ab') and F(ab')₂ fragments, single-chain
12 antibodies, recombinant immunoglobulin variable regions (Fv)s, fusion proteins etc. It is well
13 recognized in the art that some amino acid sequence can be varied in a polypeptide without
14 significant effect on the structure or function of the protein. In the molecular rearrangement of
15 antibodies, modifications in the nucleic or amino acid sequence of the backbone region can
16 generally be tolerated. These include, but are not limited to, substitutions (preferred are
17 conservative substitutions), deletions or additions. Furthermore, it is within the purview of this
18 invention to conjugate standard chemotherapeutic modalities, e.g. radionuclides, with the
19 CDMAB of the instant invention, thereby focusing the use of said chemotherapeutics. The
20 CDMAB can also be conjugated to toxins, cytotoxic moieties, enzymes e.g. biotin conjugated
21 enzymes, or hematogenous cells.

1 The prospect of individualized anti-cancer treatment will bring about a change in the
2 way a patient is managed. A likely clinical scenario is that a tumor sample is obtained at the
3 time of presentation, and banked. From this sample, the tumor can be typed from a panel of
4 pre-existing cancerous disease modifying antibodies. The patient will be conventionally
5 staged but the available antibodies can be of use in further staging the patient. The patient can
6 be treated immediately with the existing antibodies and/or a panel of antibodies specific to the
7 tumor can be produced either using the methods outlined herein or through the use of phage
8 display libraries in conjunction with the screening methods herein disclosed. All the
9 antibodies generated will be added to the library of anti-cancer antibodies since there is a
10 possibility that other tumors can bear some of the same epitopes as the one that is being
11 treated. The antibodies produced according to this method may be useful to treat cancerous
12 disease in any number of patients who have cancers that bind to these antibodies.

13 Using substantially the process of US 6,180,370, the mouse monoclonal antibody
14 11BD-2E11-2 was obtained following immunization of mice with cells from a patient's breast
15 tumor biopsy. Within the context of this application, anti-cancer antibodies having either cell-
16 killing (cytotoxic) or cell-growth inhibiting (cytostatic) properties will hereafter be referred to
17 as cytotoxic. This antibody can be used in aid of staging and diagnosis of a cancer, and can be
18 used to treat tumor metastases. The 11BD-2E11-2 antigen was expressed on the cell surface of
19 a broad range of human cell lines from different tissue origins. The breast cancer cell line
20 MCF-7 and ovarian cancer cell line OVCAR-3 were the only two cancer cell lines tested that
21 were susceptible to the cytotoxic effects of 11BD-2E11-2 as described in S/N 10/348,231.

1 The *in vitro* effects of 11BD-2E11-2 against breast and ovarian cancer cells were
2 extended by establishing its anti-tumor activity *in vivo*. *In vivo* models of human cancer were
3 established by implanting the MCF-7 breast cancer cells or OVCAR-3 ovarian cancer cells
4 into severe combined immunodeficient (SCID) mice, as they are incapable of rejecting the
5 human tumor cells due to a lack of certain immune cells. The effects of drugs tested in these
6 kinds of pre-clinical xenograft tumor models are considered valid predictors of therapeutic
7 efficacy. Cancer xenografts in mice grow as solid tumors developing parenchyma, stroma,
8 central necrosis and neo-vasculature in the same manner as naturally occurring cancers. The
9 mammary cancer cell line MCF-7 and the ovarian cancer cell line OVCAR-3 have been
10 evaluated in SCID mice. The successful engraftment of both the MCF-7 and OVCAR-3
11 tumors and the sensitivity of the tumors to standard chemotherapeutic agents have
12 characterized them as suitable models of human cancer for drug testing. The MCF-7 parental
13 cell line and its variants and the OVCAR-3 cell line have been used successfully in xenograft
14 tumor models to evaluate a wide range of therapeutic agents that have been used as clinical
15 chemotherapeutic agents.

16 11BD-2E11-2 prevented tumor growth and reduced tumor burden in a preventative *in*
17 *vivo* model of human breast cancer. Monitoring continued past 280 days post-treatment. 40
18 percent of the 11BD-2E11-2 treatment group was still alive at over 7.5 months post-
19 implantation. Conversely, the isotype control group had 100 percent mortality after 6.5
20 months post-treatment. At day 51 (soon after last treatment), the mean tumor volume in the
21 11BD-2E11-2 treated group was 20% of the isotype control (p=0.0098). Therefore 11BD-
22 2E11-2 enhanced and decreased the tumor burden compared to the control-treated groups in a

1 well-established model of human breast cancer.

2 In addition to the beneficial effects in a model of human breast cancer, 11BD-2E11-2
3 treatment also had anti-tumor activity against OVCAR-3 cells in an ovarian cancer model.
4 Body weight was used a surrogate measure of tumor progression in this model. At day 80 post-
5 implantation (16 days after the end of treatment) the mice in the treated group had 87.6 percent
6 the mean body weight of the control group ($p=0.015$). Thus, 11BD-2E11-2 treatment was
7 efficacious as it delayed tumor progression compared to the buffer control treated group in a
8 well-established model of human ovarian cancer. The anti-tumor activities of 11BD-2E11-2,
9 in several different cancer models, make it an attractive anti-cancer therapeutic agent.

10 In all, this invention teaches the use of the 11BD-2E11-2 antigen as a target for a
11 therapeutic agent, that when administered can reduce the tumor burden of a cancer expressing
12 the antigen in a mammal, and can also lead to a prolonged survival of the treated mammal.
13 This invention also teaches the use of CDMAB (11BD-2E11-2), and its derivatives, to target
14 its antigen to reduce the tumor burden of a cancer expressing the antigen in a mammal, and to
15 prolong the survival of a mammal bearing tumors that express this antigen.

16 If a patient is refractory to the initial course of therapy or metastases develop, the
17 process of generating specific antibodies to the tumor can be repeated for re-treatment.
18 Furthermore, the anti-cancer antibodies can be conjugated to red blood cells obtained from that
19 patient and re-infused for treatment of metastases. There have been few effective treatments
20 for metastatic cancer and metastases usually portend a poor outcome resulting in death.
21 However, metastatic cancers are usually well vascularized and the delivery of anti-cancer
22 antibodies by red blood cells can have the effect of concentrating the antibodies at the site of

1 the tumor. Even prior to metastases, most cancer cells are dependent on the host's blood
2 supply for their survival and an anti-cancer antibody conjugated to red blood cells can be
3 effective against *in situ* tumors as well. Alternatively, the antibodies may be conjugated to
4 other hematogenous cells, e.g. lymphocytes, macrophages, monocytes, natural killer cells, etc.

5 There are five classes of antibodies and each is associated with a function that is
6 conferred by its heavy chain. It is generally thought that cancer cell killing by naked
7 antibodies are mediated either through antibody-dependent cell-mediated cytotoxicity (ADCC)
8 or complement-dependent cytotoxicity (CDC). For example murine IgM and IgG2a antibodies
9 can activate human complement by binding the C-1 component of the complement system
10 thereby activating the classical pathway of complement activation which can lead to tumor
11 lysis. For human antibodies, the most effective complement activating antibodies are generally
12 IgM and IgG1. Murine antibodies of the IgG2a and IgG3 isotype are effective at recruiting
13 cytotoxic cells that have Fc receptors which will lead to cell killing by monocytes,
14 macrophages, granulocytes and certain lymphocytes. Human antibodies of both the IgG1 and
15 IgG3 isotype mediate ADCC.

16 Another possible mechanism of antibody mediated cancer killing may be through the
17 use of antibodies that function to catalyze the hydrolysis of various chemical bonds in the cell
18 membrane and its associated glycoproteins or glycolipids, so-called catalytic antibodies.

19 There are two additional mechanisms of antibody mediated cancer cell killing which
20 are more widely accepted. The first is the use of antibodies as a vaccine to induce the body to
21 produce an immune response against the putative antigen that resides on the cancer cell. The

1 second is the use of antibodies to target growth receptors and interfere with their function or to
2 down regulate that receptor so that effectively its function is lost.

3 The clinical utility of a cancer drug is based on the benefit of the drug under an
4 acceptable risk profile to the patient. In cancer therapy survival has generally been the most
5 sought after benefit, however there are a number of other well-recognized benefits in addition
6 to prolonging life. These other benefits, where treatment does not adversely affect survival,
7 include symptom palliation, protection against adverse events, prolongation in time to
8 recurrence or disease-free survival, and prolongation in time to progression. These criteria are
9 generally accepted and regulatory bodies such as the U.S. Food and Drug Administration
10 (F.D.A.) approve drugs that produce these benefits (Hirschfeld et al. Critical Reviews in
11 Oncology/Hematology 42:137-143 2002). In addition to these criteria it is well-recognized that
12 there are other endpoints that may presage these types of benefits. In part, the accelerated
13 approval process granted by the U.S. F.D.A. acknowledges that there are surrogates that will
14 likely predict patient benefit. As of year-end (2003), there has been sixteen drugs approved
15 under this process, and of these, four have gone on to full approval, i.e., follow-up studies have
16 demonstrated direct patient benefit as predicted by surrogate endpoints. One important
17 endpoint for determining drug effects in solid tumors is the assessment of tumor burden by
18 measuring response to treatment (Therasse et al. Journal of the National Cancer Institute
19 92(3):205-216 2000). The clinical criteria (RECIST criteria) for such evaluation have been
20 promulgated by Response Evaluation Criteria in Solid Tumors Working Group, a group of
21 international experts in cancer. Drugs with a demonstrated effect on tumor burden, as shown
22 by objective responses according to RECIST criteria, in comparison to the appropriate control

1 group tend to, ultimately, produce direct patient benefit. In the pre-clinical setting tumor
2 burden is generally more straightforward to assess and document. In that pre-clinical studies
3 can be translated to the clinical setting, drugs that produce prolonged survival in pre-clinical
4 models have the greatest anticipated clinical utility. Analogous to producing positive
5 responses to clinical treatment, drugs that reduce tumor burden in the pre-clinical setting may
6 also have significant direct impact on the disease. Although prolongation of survival is the
7 most sought after clinical outcome from cancer drug treatment, there are other benefits that
8 have clinical utility and it is clear that tumor burden reduction can also lead to direct benefits
9 and have clinical impact (Eckhardt et al. Developmental Therapeutics: Successes and Failures
10 of Clinical Trial Designs of Targeted Compounds; ASCO Educational Book, 39th Annual
11 Meeting, 2003, pages 209-219).

12 Accordingly, it is an objective of the invention to utilize a method for producing
13 CDMAB from cells derived from a particular individual which are cytotoxic with respect to
14 cancer cells while simultaneously being relatively non-toxic to non-cancerous cells, in order to
15 isolate hybridoma cell lines and the corresponding isolated monoclonal antibodies and antigen
16 binding fragments thereof for which said hybridoma cell lines are encoded.

17 It is an additional objective of the invention to teach CDMAB and antigen binding
18 fragments thereof.

19 It is a further objective of the instant invention to produce CDMAB whose cytotoxicity
20 is mediated through ADCC.

21 It is yet an additional objective of the instant invention to produce CDMAB whose

1 cytotoxicity is mediated through CDC.

2 It is still a further objective of the instant invention to produce CDMAB whose
3 cytotoxicity is a function of their ability to catalyze hydrolysis of cellular chemical bonds.

4 A still further objective of the instant invention is to produce CDMAB which are useful
5 in a binding assay for the diagnosis, prognosis, and monitoring of cancer.

6 Other objects and advantages of this invention will become apparent from the
7 following description wherein are set forth, by way of illustration and example, certain
8 embodiments of this invention.

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10 BRIEF DESCRIPTION OF THE FIGURES

11 Figure 1. Effect of 11BD-2E11-2 on tumor growth in a preventative MCF-7 breast cancer
12 model. The dashed line indicates the period during which the antibody was administered. Data
13 points represent the mean \pm SEM.

14 Figure 2. Survival of tumor-bearing mice after treatment with 11BD-2E11-2 or isotype control
15 antibody in a preventative MCF-7 xenograft study. Mice were monitored for survival for
16 longer than 230 days post-treatment.

17 Figure 3. Effect of 11BD-2E11-2 on mean body weight in a preventative OVCAR-3 ovarian
18 cancer model. The solid line indicates the period during which the antibody was administered.
19 Data points represent the mean \pm SEM.

20 Figure 4. Survival of tumor-bearing mice after treatment with 11BD-2E11-2 or buffer control
21 antibody in a preventative OVCAR-3 study. Mice were monitored for survival for

1 approximately 60 days post-treatment.

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3 DETAILED DESCRIPTION OF THE INVENTION

4 EXAMPLE 1

5 The hybridoma cell line 11BD-2E11-2 was deposited, in accordance with the Budapest
6 Treaty, with the American Type Culture Collection, 10801 University Blvd., Manassas, VA
7 20110-2209 on November 11th, 2003, under Accession Number PTA-5643. In accordance
8 with 37 CFR 1.808, the depositors assure that all restrictions imposed on the availability to the
9 public of the deposited materials will be irrevocably removed upon the granting of a patent.

10 11BD-2E11-2 monoclonal antibody was produced by culturing the hybridomas in CL-
11 1000 flasks (BD Biosciences, Oakville, ON) with collections and reseeded occurring
12 twice/week and with purification according to standard antibody purification procedures with
13 Protein G Sepharose 4 Fast Flow (Amersham Biosciences, Baie d'Urfé, QC).

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15 *In Vivo* MCF-7 Preventative Survival Tumor Experiment

16 With reference to Figures 1 and 2, 4 to 8 week old female SCID mice were implanted
17 with 5 million MCF-7 human breast cancer cells in 100 microlitres saline injected
18 subcutaneously in the scruff of the neck. The mice were randomly divided into 2 treatment
19 groups of 11-13 mice. On the day after implantation, 20 mg/kg of either 11BD-2E11-2 test
20 antibody or isotype control antibody (known not to bind MCF-7 or OVCAR-3 cells) was
21 administered intraperitoneally at a volume of 300 microliters after dilution from the stock

1 concentration with a diluent that contained 2.7 mM KCl, 1 mM KH₂PO₄, 137 mM NaCl and
2 20 mM Na₂HPO₄. The antibodies were then administered once per week for a period of 7
3 weeks in the same fashion. Tumor growth was measured about every seventh day with calipers
4 for up to 8 weeks or until individual animals reached the Canadian Council for Animal Care
5 (CCAC) end-points. Body weights of the animals were recorded for the duration of the study.
6 At the end of the study all animals were euthanised according to CCAC guidelines.

7 11BD-2E11-2 significantly reduced the tumor burden in treated mice in comparison to
8 controls (Figure 1). After treatment (day 51), 11BD-2E11-2 prevented tumor growth by 80
9 percent (p=0.0098) in comparison to isotype control antibody treated mice. There was also a
10 post-treatment survival benefit (Figure 2) associated with 11BD-2E11-2 administration. The
11 isotype control antibody treated group reached 100 percent mortality by day 197 post-treatment
12 while 40 percent of the 11BD-2E11-2 treated group were still alive at day 233. In summary,
13 11BD-2E11-2 increased survival and decreased tumor burden in a well-established model of
14 human breast cancer (Blakey et al. Clinical Cancer Research 8:1974-1983 2002; Klement et
15 al. Clinical Cancer Research 8:221-232 2002; Waud et al. Relevance of Tumor Models for
16 Anticancer Drug Development, Fiebig and Burger, eds. 54:305-315 1999; Karpanen et al.
17 Cancer Research 61:1786-1790 2001).

18 19 EXAMPLE 2

20 *In Vivo* OVCAR-3 Preventative Tumor Experiments

21 With reference to the data shown in Figures 3 and 4, 4 to 8 week old, female SCID
22 mice were implanted with 5 million OVCAR-3 human ovarian cancer cells in 1000 microliters

1 saline injected intraperitoneally. The mice were randomly divided into 2 treatment groups of
2 10. On the day after implantation, 20 mg/kg of 11BD-2E11-2 test antibody or buffer control
3 antibody was administered intraperitoneally at a volume of 300 microliters after dilution from
4 the stock concentration with a diluent that contained 2.7 mM KCl, 1 mM KH₂PO₄, 137 mM
5 NaCl and 20 mM Na₂HPO₄. The antibodies were then administered once per week for a period
6 of 9 weeks in the same fashion. Body weights of the animals were recorded for the duration of
7 the study. At the end of the study all animals were euthanised according to CCAC guidelines.

8 In the OVCAR-3 ovarian cancer xenograft model, increasing body weight can be used
9 as a surrogate indicator of disease progression since this reflects the accumulation of ascites
10 from increased tumor burden (Figure 3). At day 80 post-implantation (16 days after the end of
11 treatment), 11BD-2E11-2 administration prevented body weight gain by 12.4 percent
12 (p=0.015) compared to the buffer control group. Mice were monitored post-treatment for
13 survival (Figure 4). By day 87, the buffer control group had reached 90 percent mortality
14 while the 11BD-2E11-2 treated group still had 80 percent survival. The 11BD-2E11-2 treated
15 group did not reach 90 percent mortality until day 125. In summary, 11BD-2E11-2 antibody
16 treatment reduced tumor burden, delayed disease progression and enhanced survival in
17 comparison to a buffer control antibody in a well-recognized model of human ovarian cancer.
18 Therefore treatment with 11BD-2E11-2 significantly decreased the tumor burden of
19 established tumors in two well-recognized models of human cancer disease (breast and ovarian
20 cancers) suggesting pharmacologic and pharmaceutical benefits of this antibody for therapy in
21 other mammals, including man (Smith et al. The Prostate 48:47-53 2001; Olson et al.
22 International Journal of Cancer 98:923-929 2002; Guilbaud et al. Clinical Cancer Research

1 7:2573-2580 2001; Von Gruenigen et al. International Journal of Gynecologic Cancer 9:365-
2 372 1999; Guichard et al. Clinical Cancer Research 7:3222-3228 2001; Xiao et al. Protein
3 Expression and Purification 19:12-21 2000).

4 All patents and publications mentioned in this specification are indicative of the levels
5 of those skilled in the art to which the invention pertains. All patents and publications are
6 herein incorporated by reference to the same extent as if each individual publication was
7 specifically and individually indicated to be incorporated by reference.

8 It is to be understood that while a certain form of the invention is illustrated, it is not to
9 be limited to the specific form or arrangement of parts herein described and shown. It will be
10 apparent to those skilled in the art that various changes may be made without departing from
11 the scope of the invention and the invention is not to be considered limited to what is shown
12 and described in the specification. One skilled in the art will readily appreciate that the present
13 invention is well adapted to carry out the objects and obtain the ends and advantages
14 mentioned, as well as those inherent therein. Any oligonucleotides, peptides, polypeptides,
15 biologically related compounds, methods, procedures and techniques described herein are
16 presently representative of the preferred embodiments, are intended to be exemplary and are
17 not intended as limitations on the scope. Changes therein and other uses will occur to those
18 skilled in the art which are encompassed within the spirit of the invention and are defined by
19 the scope of the appended claims. Although the invention has been described in connection
20 with specific preferred embodiments, it should be understood that the invention as claimed
21 should not be unduly limited to such specific embodiments. Indeed, various modifications of
22 the described modes for carrying out the invention which are obvious to those skilled in the art

1 are intended to be within the scope of the following claims.

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